COMPOSITION AND METHODS FOR PREDICTING NECROTIZING ENTEROCOLITIS IN PRETERM INFANTS

STATEMENT OF FEDERALLY SPONSORED RESEARCH AND DEVELOPMENT

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BACKGROUND OF INVENTION

[0002] The colonization of animal intestinal tracts with microorganisms starts at birth and undergoes rapid shifts in composition and structure as the host matures over time (Koenig et al., 2011; Sharon et al., 2013; Grier et al., 2017; de Muinck and Trosvik, 2018). These microorganisms perform essential functions mechanistically linked to the immune system development, nutrient acquisition and energy regulation, opportunistic pathogens suppression, as well as intestinal barrier competency, which includes epithelial metabolism, proliferation and survival, mucin and antimicrobial compound production, and cell-cell communication signaling molecule secretion (Neish, 2009; Belkaid and Hand, 2014; Yu et al., 2016).

[0003] Disruption of the intestinal microbiota leads to dysbiosis, a state of ecological imbalance where the community loses diversity, key bacterial species, and more critically metabolic capacity with reduced colonization resistance to opportunistic pathogens (Arrieta et al., 2014). Early life gut dysbiosis is associated with disease susceptibility along with short-term and lifelong health issues, such as necrotizing enterocolitis (Madan et al., 2012), sepsis (Madan et al., 2012), asthma and allergies (Arrieta et al., 2015), type 1 diabetes (Vatanen et al., 2016), celiac disease (Cenit et al., 2015), inflammatory bowel disease (Gevers et al., 2014) and obesity (Cho et al., 2012), among others.

[0004] In particular, necrotizing enterocolitis (NEC) is a life-threatening, gastrointestinal emergency that affects approximately 7-10% of preterm neonates with mortality as high as 30-50% (Guner et al., 2009). In this condition, bacteria cross the intestinal wall leading to local and systemic infection and inflammation, and bowel wall necrosis and perforation. Intestinal barrier immaturity, characterized as elevated intestinal permeability, or "leaky gut," is the proximate cause of susceptibility to NEC in preterm neonates (Anand et al., 2007; Fitzgibbons et al., 2009; Fox and Godavitarne, 2012; Bergmann et al., 2013).

[0005] It is critical to characterize the preterm infant intestinal microbiota to identify dysbiotic states associated with increased intestinal leakiness, as well as beneficial bacteria associated with improved intestinal barrier function, for subsequent stratification of early diagnosis, early intervention and primary prevention of leaky gut and its sequelae. The present invention is directed to these ends and to other important goals.

BRIEF SUMMARY OF INVENTION

[0006] As disclosed herein, the impact of intestinal microbiota development on intestinal mucosal barrier maturation was evaluated in preterm neonates. As discussed in detailed below, a cohort of preterm infants<33 weeks gestation was monitored for intestinal permeability (IP) and fecal microbiota during the first two weeks of life. Rapid decrease in IP indicating intestinal barrier function maturation correlated with significant increase in gut bacteria community diversity. In particular, members of the orders Clostridiales and Bifidobacteriales were highly transcriptionally active, and progressively increasing abundance of Clostridiales was significantly associated with decreased intestinal permeability, an indication of epithelial barrier maturation. Further, neonatal factors previously identified to promote intestinal barrier maturation, including early exclusive breastmilk feeding and shorter duration antibiotic exposure, were found to associate with the early colonization of the intestinal microbiota by members of the Clostridiales, which altogether are associated with improved intestinal barrier function in preterm infants. Given that intestinal barrier immaturity, or "leaky gut," is the proximate cause of susceptibility to necrotizing enterocolitis (NEC) in preterm neonates, these findings form the basis of the invention disclosed herein.

[0007] For example, and in a first embodiment, the invention is directed to a method of characterizing intestinal permeability (IP) in a subject.

[0008] In a first aspect, this method comprises determining the amount of Clostridiales and/or Bifidobacteriales bacteria in a sample obtained from a subject, wherein when the amount of Clostridiales and/or Bifidobacteriales bacteria is about 5% or less by relative abundance of the total amount of bacteria in the sample, the IP of the subject is characterized as high, and wherein when the amount of Clostridiales and/or Bifidobacteriales bacteria is more than about 5% by relative abundance of the total amount of bacteria in the sample, the IP of the subject is characterized as low. In particular aspects, the subject may be a preterm infant. In particular aspects, the sample is a stool sample.

[0009] In a second embodiment, the present invention is directed to a method of treating or preventing high intestinal permeability in a subject.

[0010] In a first aspect, this method comprises determining the amount of Clostridiales and/or Bifidobacteriales bacteria in a sample obtained from a subject, and administering a therapeutically effective amount of a treatment or preventive agent for high intestinal permeability to the subject when the amount of Clostridiales and/or Bifidobacteriales bacteria in the sample is about 5% or less by relative abundance of the total amount of bacteria. In particular aspects, the subject may be a preterm infant. In particular aspects, the sample is a stool sample.

[0011] In a second aspect, this method comprises determining the amount of Clostridiales and/or Bifidobacteriales bacteria in a sample obtained from a subject, and administering a therapeutically effective amount of a treatment or preventive agent for high intestinal permeability to the subject when the amount of Clostridiales and/or Bifidobacteriales bacteria is within a pre-established range of amounts of Clostridiales and/or Bifidobacteriales bacteria associated with high intestinal permeability. In particular aspects, the subject may be a preterm infant. In particular aspects, the sample is a stool sample.

[0012] In a third aspect, this method comprises determining the amount of Clostridiales and/or Bifidobacteriales bacteria in samples obtained from a subject at two or more time points and administering a therapeutically effective amount of a treatment or preventive agent for high intestinal permeability to the subject when the amount of Clostridiales and/or Bifidobacteriales bacteria in the samples decreases